

Proposed Path Forward

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Purpose

- Discuss a proposed path forward
- Goal: successfully enhancing the bioremediation of dissolved benzene through the addition of sulfate; however, it is not expected to be effective in areas with LNAPL

So, where can we go from here?

- Aerobic benzene biodegradation pathways well known, identified
- But anaerobic benzene biodegradation pathways are not well known
- So, what can be done to maximize chance of success, for biodegrading benzene in groundwater, when sulfate is added?
 - Possible Idea: Bioaugmentation

Bioaugmentation

- Can help provide necessary microbes to an environment that lacks the needed population
- Can help provide necessary microbes if needed population is present but of a low size and needs time to grow
 - Can prevent long lag-time (which otherwise can possibly be years) after nutrient addition
- Bioaugmentation coupled with sulfate addition
 - Can be performed with flexible timing, including after sulfate addition
 - Can help drive benzene biodegradation in presence of sulfate
- When dissolved benzene in groundwater is the target, will still have to contend with the preference for indigenous microbes to target biodegradation of toluene, ethyl-benzene, and xylenes

Example: Delta proteobacterium ORM2

- Biodegrades benzene under sulfate-reducing and methanogenic conditions
 - Unknown mechanism
- Quant Array (Petroleum) test does not analyze for it, but there is a commercially-available qPCR test available to quantify it
- Testing involves simple grab sample of GW
- 48-72 hour turnaround time for results

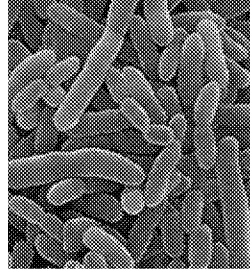


Image: E. Edwards et al, 2017

Example: Delta proteobacterium ORM2

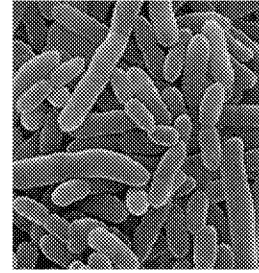


Image: E. Edwards et al, 2017

- Potential outcomes of ORM2 quantification
 - Population may be present, is a healthy size
 - Population may not be present, or size may be small
 - Population may be present and healthy in some locations but not all
- ORM2 bioaugmentation can be used to support EBR, help successfully drive anaerobic, dissolved-benzene bioattenuation forward
 - Bioaugmentation with these organisms has not yet been tested in the field, the cultures are not available commercially, but pilot test sites are being sought
 - These cultures may not be compatible with environmental conditions throughout the contaminated area of ST-12
 - These cultures may not be compatible with planned sulfate injection concentrations, but bench testing could be done to evaluate this
 - Documentation of success of the bioaugmentation process will require significantly increased monitoring network

Summary

- Currently available microbial data has not determined if the bioremediation of dissolved benzene in groundwater will be enhanced with just the addition of sulfate
- A proposed path forward includes:
 - Performing an assessment for the presence of ORM2
 - Determine if bioaugmentation with ORM2 will be helpful in meeting site goals
- The goal is to maximize the chance for EBR success after sulfate additions to the site

Additional Slides in Case of Questions

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In Search of Elusive Anaerobic Benzene Activation Mechanisms

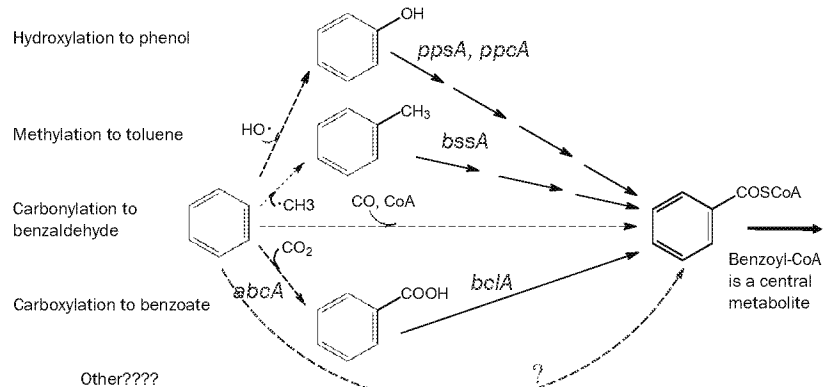


Image: E. Edwards, 2017 3

In Search of Elusive Anaerobic Benzene Activation Mechanisms

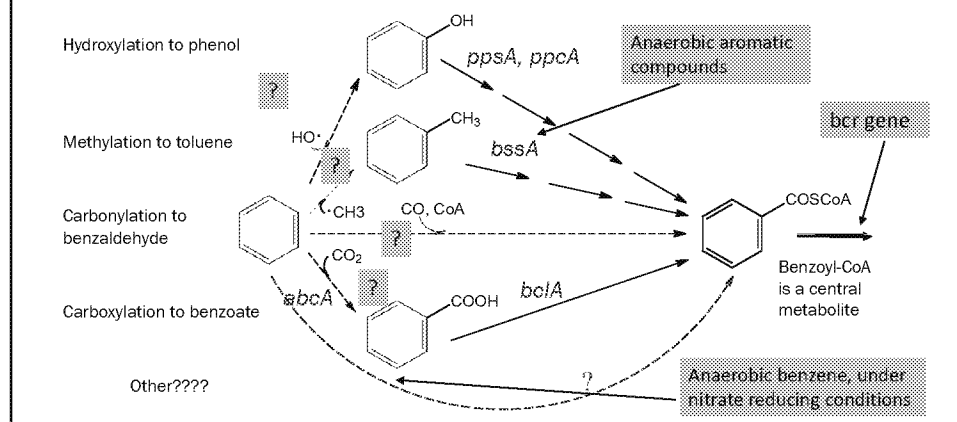
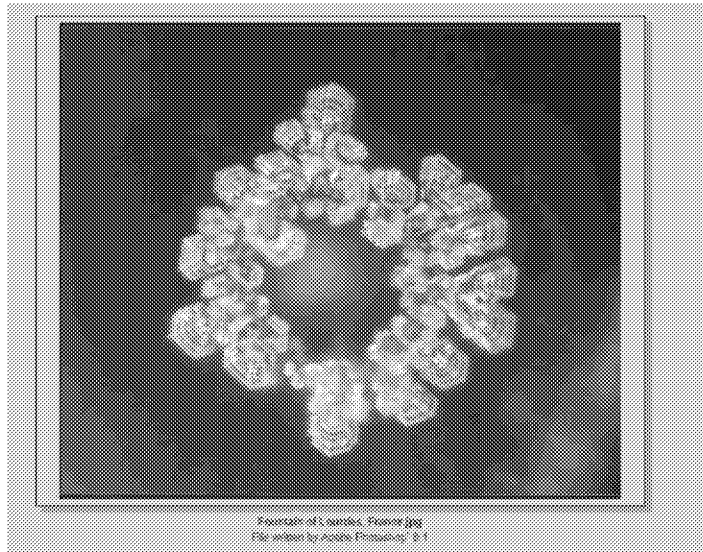


Image modified from E. Edwards, 2017

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Freezing of 1. water, 1. water 1.1
File written by Masaru Emoto, 1.1

<http://www.masaru-emoto.net/english/water-crystal.html>